REMARKS

Claim Amendments

Claims 1, 2, 4, 5, 9-11, 30, 31, 33, 35, 37, 48, 52, 55, and 60-66 are in the application. Claim 55 is amended. New claims 67-84 are added.

Claim 55 is amended to correct antecedent basis in the ischemic renal tubular cell injury.

New independent Claim 67 is based on Claim 1 as originally filed, wherein the renal tubular cell injury (RTCI) is selected from an ischemic renal injury; to provide that the urine sample is obtained within twenty-four hours of the RTCI [express support in para. 0039]; to provide that the mammal is suspected of having the RTCI [implicit support throughout the description and figures, including in para. 0037 and 0043]; and to provide "correlating the level of detected antibody-NGAL complex to the mammal having the RTCI [express support in para. 0043].

New independent Claim 68 is based on Claim 1 as originally filed, wherein the renal tubular cell injury is the ischemic renal injury, the mammal is a human patient, the steps (a) and (b) of originally-filed Claim 1 have been combined into a single step, and the biomarker consisting of NGAL.

New Claims 69-71 are supported by Claims 2 - 4 as originally filed, respectively.

New Claim 72 is based on Claim 5 as original filed, made dependent upon Claim 68, and by combining the steps 1) and 2) of originally-filed Claim 5, and step 2) of originally-filed Claim 1, into a single step 3).

New Claims 73 and 74 are supported by Claims 10 and 11 as originally filed, respectively.

New Claim 75 is based on Claim 28 as original filed, made dependent upon Claim 68, and provides that the urine sample comprises up to 1 milliliter of the first urine from the patient.

New Claim 76 depends from Claim 68 and provides for the detection of ischemic renal injury in human patients after kidney transplantation, wherein the urinary NGAL measured within two hours of kidney transplantation is predictive of acute renal failure, and is supported expressly by paragraph [0100].

New Claim 77 depends from Claim 68 and provides for the detection of post-operative acute renal failure in human patients after open heart surgery, wherein the urinary NGAL

measured within two hours after surgery is predictive of acute renal failure, and is supported by paragraph [0101] and Figure 16.

New Claim 78 depends from Claim 77 and provides that patients who subsequently develop acute renal failure display a greater than 10-fold increase in the 2 hour value for urinary NGAL expressed as ng NGAL/mg creatinine, as compared to those who do not, and is supported by paragraph [0101].

New Claim 79 depends from Claim 77 and provides that patients who subsequently develop acute renal failure display a greater than 20-fold increase in the 4 hour value for urinary NGAL expressed as ng NGAL/mg creatinine, as compared to those who do not, and is supported by paragraph [0101].

New Claims 80 and 81 depend from Claim 68, and provide that urine sample is within four hours and two hours, respectively, following the renal tubular cell injury, and are supported in paragraph [0051].

New independent Claim 82 is based on Claim 24 as originally filed, wherein the renal tubular cell injury is an ischemic renal injury, and further provides that NGAL appears within the first 24 hours of the onset of the ischemic renal injury, which is supported in paragraph [0039].

New claims 83 and 84 depend from Claim 82 and are supported by Claims 26 and 27 as originally filed, respectively.

Applicants believe the amendments to the claims find full support in the specification, and that no additional claim fees are due.

Priority

Applicants understand that the Office recognizes the claim of priority to provisional applications 60/458,143 and 60/481,596, and that the effective filing dates of independent claim 66 and dependent claims 2, 4-5, 9-11, 33, 35 and 55 therefrom, is March 27, 2003.

The rejection denies any benefit of priority to the provisional applications for claims 37 and 60, and recognizes an effective filing date of March 29, 2004 for claims 37 and 60.

Applicants assert that new Claims 67-75 and 80-84 have express support in both provisional applications 60/458,143 and 60/481,596, and mirror the claims granted in the corresponding EPO application of the present invention.

Claim Objection

Applicants thank the examiner for her kindness in pointing out this objection. Claim 55 has been amended to obviate the objection.

Claim Rejections

Claim 60 is rejected under 35 U.S.C. 112, second paragraph, as being indefinite.

Applicants request reconsideration and withdrawal of the rejection in view of the amendment made to Claim 60, providing that the urine sample is obtained at 2 hours after surgery.

Claims 4-5, 9-11, 33, 35, 37, 55 and 66 are rejected under 35 U.S.C. 103(a) as being unpatentable over either "Matthaeus 1" or "Matthaeus 2", in view of "Ramsden", "Blaser", "Moses", "David", and "Muramatsu"; or, in the alternative, over either "Matthaeus 1" or "Matthaeus 2" and "Ohlsson", in view of "Ramsden", "Blaser", "Moses", "David", and "Muramatsu".

Applicants traverse.

When making an obviousness rejection, Office personnel must ensure that the written record includes findings of fact concerning the state of the art and the teachings of the references applied. In certain circumstances, it may also be important to include explicit findings as to how a person of ordinary skill would have understood prior art teachings, or what a person of ordinary skill would have known or could have done. Factual findings made by Office personnel are the necessary underpinnings to establish obviousness. The focus when making a determination of obviousness should be on what a person of ordinary skill in the pertinent art would have known at the time of the invention, and on what such a person would have reasonably expected to have been able to do in view of that knowledge. The prior art reference (or references when combined) need not teach or suggest all the claim limitations, however, Office personnel must explain why the difference(s) between the prior art and the claimed

invention would have been obvious to one of ordinary skill in the art. The gap between the prior art and the claimed invention may not be "so great as to render the [claim] nonobvious to one reasonably skilled in the art." (MPEP 2141).

Applicants at the outset assert that the rejection is based on inaccurate and incomplete findings of fact concerning the teaching of the Matthacus and other references, and that a *prima facie* obviousness rejection necessarily cannot stand if it is based upon inaccurate or incomplete findings of fact.

The rejection at page 7 lines 3-9, first recites that "Matthaeus 1 teach that levels of NGAL protein are upregulated in response to experimentally induced acute ischemic renal injury in a rat model (i.e., ischemic renal tubular cell injury; see entire selection). By contrast, control animals displayed only minor expression of NGAL, demonstrating that renal injury and repair is associated with an upregulation of NGAL (i.e., correlating the level of NGAL with ischemic renal tubular cell injury). Matthaeus 1 further state that NGAL was elevated "after 24 and 48 hours" of renal ischemia as assessed by Western blot analysis."

The rejection at page 7 line 20 to pg 8 line 4, also states that the "Matthaeus references make clear that their studies were performed on rats *as an animal model of human disease* (this is made explicit in Matthaeus 2, who refer to a "rat model of renal ischemia"). Matthaeus 1 state that the purpose of their experiment is to "further elucidate the processes involved in renal injury and repair". The findings reported therein support a "critical role in the renal response to injury" for NGAL, correlating upregulation of NGAL with ischemic renal tubular cell injury."

(a) Matthaeus teaches an association between NGAL and MMP-9 in the post-ischemic kidney

Applicants disagree with this characterization of Matthaeus 1 and Matthaeus 2. (Hereinafter, Matthaeus 1 and Matthaeus 2 will be referred to collectively as "Matthaeus" since their disclosures are nearly identical). The Applicants in the accompanying 132 Declaration of inventors Barasch and Devarajan (hereinafter, "Barasch/Devarajan 132 Declaration") at paragraph 8-11, correct the factual finding made in the rejection by emphasizing that Matthaeus focused on an association of MMP-9 with NGAL. Matthaeus stated "(w)e conclude that MMP-9 and NGAL may play a critical role in the renal response to ischemic injury." (emphasis added)

Matthaeus repeatedly mentions the association of MMP-9 and NGAL; specifically, "NGAL has been shown to occur in disulfite-linked complexes with matrix metalloproteinase-9 (MMP-9) and tissue inhibitor of metalloproteinase-9 (TIMP-1)"; "(e)xpression of NGAL and its associated molecules, MMP-9 and TIMP-1, was studied"; and "(a)s MMP-9 and NGAL are simultaneously upregulated in injured proximal tubuli...".

The rejection makes fact findings from the teaching of Matthaeus, but selectively in terms of only NGAL and its alleged role, ignoring its association with MMP-9. Applicants state in the Barasch/Devarajan 132 Declaration at paragraph 9 that they do not believe that Matthaeus' emphasis on an association between MMP-9 and NGAL can be so easily trivialized by focusing only on NGAL and its role, pointing out that at the time of Matthaeus, it was thought that the interaction with MMP-9 was the key function of NGAL. For example, Yan et al. [J Biol Chem. 2001 Oct 5;276(40):37258-65, which is of record] showed that NGAL was capable of protecting MMP-9 from degradation in a dose-dependent manner and thereby preserving MMP-9 enzymatic activity. Yan et al. teaches that the complex was a marker of cancer, and they hypothesized that NGAL might be involved in tumor progression via its interaction with MMP-9 in view of other recent findings that showed NGAL expression levels were up-regulated in colorectal neoplasia and several epithelial carcinomas. The Applicants also emphasize the compositional difference between the NGAL:MMP-9 complex addressed in Matthaeus, as compared to the NGAL compound they found in urine, noting that the size of the NGAL reported by Yan et al. is 35,000, whereas the NGAL reported by Applicants is 25,000.

Applicants go on to state in the Barasch/Devarajan 132 Declaration at paragraph 10 and 11, that the NGAL:MMP-9 compound addressed in Matthaeus and the prior art is a cancer marker that results from chemical linkages. These chemical (disulfide linkages) are even more common in rodents than in humans (and Matthaeus used rodents) because in rodents there is an extra-unpaired cysteine residue in rodent NGAL (but not in the human) and this amino acid is involved in cross-linking. Applicants would conclude that Matthaeus is focused on the biology of MMP-9 activity in the body of the kidney, prominent in rodents. This MMP-9/NGAL complex was known to be covalently associated, and hence its separation into components would be understood to require chemical reactants. Matthaeus' description would suggest that the NGAL in the proximal tubuli was associated with MMP-9 in the kidney tissue as the MMP-9/NGAL

complex, which is not the same chemical compound as NGAL alone. Indeed, the function of NGAL in Matthaeus relates to its association with MMP-9 and its proteolytic activity.

Applicants also call to the Examiner's attention the attached 132 Declaration of Dr. Walter J. Keirans, Division Vice President of Abbott Diagnostics Division, Abbott Laboratories, Abbott Park, IL. Abbott Laboratories is the exclusive licensee of subject U.S. Application No. 10/811,130.

At page 7 in paragraph 8(a), Dr. Keirans discusses the Matthaeus references, as a person of skill in the art, stating "(t)he results set forth in abstract form in Matthaeus 1 and 2 provide insight into the appearance and changes related to biomolecules found by representational difference analysis (RDA). Experimental and control animal results are compared in which MMP-9, TIMP1 and NGAL are upregulated in kidney ischemia rat models. It should be noted by definition of RDA, the statement of function cannot be addressed in these static models, particularly when controls are not the same animals tested at all time points in the experimental protocol nor are the 12 and 24 hour animals. Numbers of animals sacrificed at each test point are not declared leaving open the statistical validity of the results. Therefore, the subsequent impact of findings in individual rats on kidney function is impossible to conclude as each animal is sacrificed at stated test points – there can be no functional outcomes. Kidney failure is extrapolated from immunohistochemistry and composite results. Moreover, the results set forth in abstract form in Matthaeus 1 and 2 appear not to have published subsequently anywhere else. Thus, it is near impossible for the applicants and Examiner to understand exactly ***what*** may or may not have been uncovered by the investigators. The lack of later publication of a full report - - unusual - - calls into question exactly what the references teach or suggest."

(b) <u>The rejection neglects to ascertain the effect of the location of NGAL expression in the kidney tissue of Matthaeus.</u>

The Barasch/Devarajan 132 Declaration at paragraph 12 corrects the rejection's inaccurate fact finding that Matthaeus teach "correlating upregulation of NGAL with ischemic renal tubular cell injury". Quite the contrary, according to Applicants, Matthaeus teach that levels of NGAL *and* MMP-9 protein are upregulated in response to experimentally induced acute ischemic renal injury in a rat model, and that renal injury and repair was associated with an upregulation of NGAL and MMP-9. The very abbreviated disclosure of Matthaeus teaches only

a single datum point of NGAL protein detection in the kidney tissue (and none whatsoever in the urine) that is made long after the initial induced injury. From this single datum, a person of ordinary skill would not conclude that the alleged upregulation of NGAL within the kidney tissue after 24 hours teaches "correlating the level of NGAL with ischemic renal tubular cell injury", as the Examiner alleges. A correlation requires a determination of the strength and direction of a relationship between two variables, and Matthaeus' very limited results fail to establish any such alleged correlation, and certainly none as between NGAL and ischemic renal tubular cell injury.

The Barasch/Devarajan 132 Declaration at paragraph 13 also notes that the rejection's fact finding of Matthaeus fails to mention, and therefore ignores the importance of, the location and timing of NGAL *and* MMP-9 expression. As will be discussed hereinafter, the actual location and timing of NGAL expression described in Matthaeus is important and essential to an accurate finding of facts. The examiner is required to make an appropriate finding of facts on the reference as a whole, and cannot ignore other material facts disclosed in Matthaeus, which would be important to a person of ordinary skill, though not conducive to forming a *prima facie* rejection (see MPEP 2141.02 (VI)). Only after a finding of the facts of the reference as a whole are presented, can the examiner properly address the question of what a person of ordinary skill would have understood and could have done.

To appreciate what a person of ordinary skill would understand concerning the location and timing of NGAL expression in Matthaeus, an understanding of the role and function of the proximal tubuli is needed. As stated at paragraph 29 of the Barasch/Devarajan 132 Declaration, the main function of the proximal renal tubules is reabsorption and degradation of proteins filtered from the circulating blood stream. The presence of NGAL protein in the proximal tubuli would indicate to a person of ordinary skill the capture of NGAL from the glomerular filtrate which is derived not from the kidney itself but from the circulating serum. Most substances that appear in the proximal tubules of the kidney do not necessarily then also show up in the urine. This is also true for NGAL. The Applicants note that Matthaeus in fact confirms this, by finding that NGAL was accumulated in the proximal tubuli, where the megalin receptors had captured and were in the process of degrading (via lysosomes) the NGAL protein, as is the normal process, and this would have then informed a person of ordinary skill in the art that the then well-known megalin process was functional. A person of ordinary skill in the art would not have

predicted that NGAL that was observed in the proximal tubuli at 24 hours would have been likewise observed in urine, and the capture of NGAL by the proximal tubuli is likely to be exclusive of any potential urinary location.

The Applicant then provide in the Barasch/Devarajan 132 Declaration, in paragraphs 30-35, a thorough discussion of state of the knowledge that includes the teachings of Brennan and Rector, Christensen et al, Nykjer et al, and Hvidberg, related to the degradation of proteins in the proximal tubules via the lysosomal pathway, and the role of megalin.

Applicants state that it is well known that proteins absorbed in the proximal tubules exclusively are degraded via the lysosomal pathway, and they thus do not pass into the urine. For example, the "The Kidney" textbook by Brenner and Rector, third edition of 1986 and its page 30, (appended hereto) gives a good description, starting on the left side, first full paragraph, of the reabsorption and degradation of plasma macromolecules and proteins ("proximal tubuli endocytosis"). Thus, proximal tubuli endocytosis is a well-known phenomenon and has been studied in detail by various researchers, including others at the time of the invention.

Applicants go on to state that Christensen in Am. J. Physiol. Renal Physiol. 280: F562-F573, 2001 has investigated the role of inter alia megalin which is one of the receptors involved in proximal tubuli endocytosis (see text starting at page F565, at bottom of left column, through page F566, right column, 2nd para). Megalin mediates the endocytosis of a wide variety of ligands of considerably differing chemical nature, structure and constitution, many of which are proteins of various sizes (see in particular table 1 on F566). Nykjer et al. in Cell, Vol.96, 507-515 (February 19, 1999) reports for example in detail on the endocytosis of vitamin D-type steroids, which is only one of the many examples given in the table 1 of Christensen. Nykjer shows that endocytosis is suppressed in megalin-knock out organisms (see Nykjer, page 512 - end of the right column).

Christensen underscores that "(m)ost, if not all of the ligands taken up by megalin or cubilin in proximal tubuli are degraded in lysosomes" (see F655, left column) and concludes that "Absence or dysfunction of either receptor is associated with significant proteinuria, showing that both are important for normal absorption of filtered protein (see F570, left column: Conclusion).

With this background, the Applicants conclude in the Barasch/Devarajan 132 Declaration that the skilled person would have also regarded NGAL susceptible to proximal tubuli

endocytosis. Indeed, a paper published by Hvidberg et al. in the Federation of European Biochemical Societies (FEBS) Letters 579, 773-777 in 2005 (appended hereto), subsequently confirms, as a result of studies conducted by a separate group of researchers in a period of time close to the filing date of Applicants'application, that NGAL is deemed prone to being strongly absorbed by megalin.

Hvidberg reports: "The other receptor is megalin [8], a multi-ligand endocytosis receptor that is expressed on a variety of epithelia, primary such that have a high adsorptive capacity such as tubular epithelia cells of kidneys..." (see page 773, right column, middle of the first paragraph); and "...thus indicating that megalin is involved in mediating the cellular uptake of NGAL by the cells"; "...internalized NGAL is not recycled but segregated from the receptor and targeted to endosomes and lysosomes." (see page 775, left column, in section 5, reporting results, last but seven lines from the end of the paragraph, as well as conclusive statement of the said paragraph.); "(w)e demonstrate here that megalin acts as such as a cellular receptor for NGAL." (see page 775, right column, in section 6, reporting discussion, at about two thirds from the beginning of the first paragraph.); and, as a conclusion: "The studies of uptake indicate that the capacity of megalin to endocytose NGAL is very high." (see page 776, left column, last sentence of the first paragraph. The Hvidberg reference comes mainly as a reported individual confirmation of the skilled man's understanding. This understanding existed in any case prior to Hvidberg.

Nothing prevents the Office from considering the Hvidberg reference the same way, merely on the grounds that it was not produced by the Applicant himself. On the contrary, independent experimental confirmation from distinct parties other than the Applicant -- as is Hvidberg -- should be granted, on the balance, even possibly an *increased* (rather than diminished) procedural weight.

Therefore, an interpretation of the prior art does not support -- at the priority date – that urine was *self-evidently* understood as "the" observation window for the NGAL reported in Matthaeus et al. Rather, what the skilled man was concerned with, was the high probability of NGAL endocytosis -- and indeed both Matthaeus 1 and Matthaeus 2 remain totally silent on NGAL's potential as a urinary biomarker of IRI -- interestingly *in spite* of the undisputed long-felt need in the art for an innovative biomarker, and the well-known problems associated with creatinine in this role.

The Applicants at paragraph 36 conclude that a person of ordinary skill in the art would not have predicted that NGAL that was observed in the proximal tubuli at 24 hours would have been likewise observed in urine, and that the skilled person would have considered that a finding of NGAL protein expression (accumulation) only in the proximal tubuli, a place where it is typically reabsorbed, suggests its absorption and degradation exclusively via the lysosomal pathway, but would not suggest its appearance in the urine. The only evidence of the ultimate outcome of such NGAL provided by Matthaeus -- that NGAL appears in the proximal tubuli -- would lead a person of skill to not expect to find the NGAL in the urine.

The Barasch/Devarajan 132 Declaration at paragraph 37 goes on to state that a contrary finding that NGAL expressed in the renal proximal tubuli then passes into the urine would have been quite a surprise to a person of ordinary skill. It has only been through the present invention that it became known that NGAL stemming from renal tubular cell injury, is readily and visibly detectable in the urine and that NGAL expression in the urine can be univocally correlated with renal tubular cell injury as the underlying cause. Applicant Prasad Devarajan states that he was surprised when he first observed that NGAL was detected in the urine following an induced renal ischemia. Examples 5 or 6 of the Applicants' specification are mentioned in support.

(c) Matthaeus lacks adequate description of the induced ischemia

Finally, the Applicants also point out in paragraph 14 of Barasch/Devarajan 132 Declaration that Matthaeus do not provide a detailed description of the technique used for inducing an ischemic renal injury, other than a "rat model of renal ischemia" and the "postischemic kidney". Matthaeus does not describe whether only one kidney (unilateral ischemia) or both kidneys (bilateral ischemia) were clamped, or for how long. The Applicants note that in other references authored by Bonventre (the last named author of Matthaeus), "postischemic kidney" refers exclusively to a unilaterally-clamped kidney (see Ichimura et al, of record) and US Patent 6,664,385 (Sanicola-Nadel et al, of record). As described in the specification (paragraph [0083]), mice with unilateral renal ischemia display serum creatinine levels that are indistinguishable from control animals, while mice with bilateral ischemia show significant elevation of serum creatinine. The Applicants state that there would be considerable uncertainty as to the extent of ischemic renal injury induced on the rat subjects of Matthaeus, and

no predictability whether such rat subjects would have exhibited elevated levels of serum creatinine that accompany acute renal failure (ARF).

(d) The rejections wrongly alleges "well-known facts"

The Office Action at page 8 lines 5-8 states that "(i)t was well known in the art that disease processes may produce changes in the levels of certain specific analytes, and that measurement of the levels of such analytes can be used to detect the presence of the disease. This is taken to be admitted prior art because applicant has failed to traverse this assertion (according to MPEP 2144.03)."

Applicants traverse the assertion that Applicants failure, if any, to traverse this statement transforms it into "admitted prior art". The same MPEP 2144.03 requires that "the examiner must provide specific factual findings predicated on sound technical and scientific reasoning to support his or her conclusion of common knowledge". As stated by Applicants in the Barasch/Devarajan 132 Declaration at paragraphs 15-16, the examiner's assertion fails if only because the "common knowledge statement" alleged to have been admitted by the Applicants, is at best a generalized principle that provides no specifics that would characterize them as facts. The statement "disease processes generally may produce changes in the levels of certain specific analytes, and that measurement of the levels of such analytes can be used to detect the presence of the disease" (emphasis added) lacks the specifics for a person of ordinary skill to either support or refute the statement, and was understood to be a generalized concept that may or may not be true depending on the specifics, which are completely lacking. The alleged statement may or may not be true, and unpredictably so, depending on the specific disease, the specific analyte, and the timing and degree of change in the level of specific analyte. The statement establishes no predictability that NGAL can be associated with an ischemic renal injury outside of the context described in Matthaeus: that is, within kidney tissue, and specifically only in the proximal tubuli, after 24 hours. Therefore, any alleged failure to have refuted such statement does not extend the scope of the statement to any specific disease or analyte in any way.

Applicants also point out that in the response filed by Applicants on November 26, 2007, to the Office Action mailed May 29, 2007 that first offered the statement "(i)t is well known in the art that disease processes may product changes in the level of certain specific analytes, and that measurement of the levels of such analytes can be used to detect the presence of the

disease", Applicants made in fact a specific traversal of the applicability of such a generalized statement as it pertained to NGAL, the urine, and biomarkers: Applicants stated: "Applicants believe that a person of ordinary skill in the art, in view of Matthaeus et al, would not have considered NGAL as a biomarker of renal injury...", and "a person of ordinary skill in the art would not find it obvious to employ Applicants' claimed sampling method in view of Matthaeus et al." (pp 37-38). Responses made to subsequent rejections that included the same, generalized, non-specific statement specifically did not acquiesce to such rejection or its basis. Such non-acquiescence cannot then transform such a non-specific generalization into "admitted prior art" under MPEP 2144.03.

(e) Matthaeus provides no teaching of a "biomarker"

The rejection goes on at page 8 lines 9-17 to state that "it would have been obvious to one of ordinary skill in the art to detect NGAL for the purpose of diagnosing acute renal injury in light of the teachings of Matthaeus that NGAL is specifically elevated in this disease condition, and further in view of the general knowledge of one skilled in the art that markers changed in response to disease can be used as biomarkers for diagnosis of the disease." (emphasis added)

The rejection attempts to rely upon a plain generalization, which may or may not be true depending upon the facts, to establish that Matthaeus et al. teaches that NGAL "could be used as a biomarker of renal ischemia, such that it would have been obvious to detect NGAL in human subjects for the clear benefit of diagnosing human disease".

Applicants traverse this conclusion of fact.

As clearly stated in Barasch/Devarajan 132 Declaration at paragraphs 18, a factual finding that Matthaeus in combination with general knowledge teach detecting NGAL for the purpose of diagnosing acute renal injury is inaccurate and baseless. Matthaeus mentions nothing whatsoever about a biomarker, and there is no teaching in Matthaeus that would enable or permit "general knowledge" to extend its teaching as such. The examiner concludes that NGAL is a biomarker for the diagnosis of ischemic renal injury in humans, solely on the basis that NGAL was found to associate with the protein MMP-9 in the post-ischemic kidney tissue. However, such a conclusion, at the very least, requires that the teaching of Matthaeus would lead a person of ordinary skill to recognize that NGAL has the properties and attributes of a biomarker. Such a recognition would be completely impossible in view of Matthaeus' teaching itself, that the

expression of NGAL is at the wrong place (proximal tubuli), at the wrong time (after 24 hours) and in the wrong form (in an association with MMP-9).

Incidentally, it is noted that indeed Matthaeus et al. do not label NGAL as a "biomarker" for injured renal proximal tubuli - which is more than understandable in view of the missing tight correlation.

Applicants note in the Barasch/Devarajan 132 Declaration at paragraph 19 that the Examiner previously has asserted by implication the requirements for a biomarker. In the Office Action dated May 29, 2007, the Examiner recognized the "laborious and lengthy nature of biomarker validation recognized by those skilled in the art, citing as examples, Bast et al. ("Translational Crossroads for Biomarkers" Clin Cancer Res 2005; 11(17),6103-6108) -- a reference that published two years after the filing date of this application -- , which points to the "lengthy process" of assay development and validation and notes that many markers that correlate with disease statistically may not prove to be useful clinically (p. 6105, right column). See also LaBaer et al. ("So, You Want to Look for Biomarkers" Journal of Proteome Research 2005; 4, 1053-1059), which teaches that crucial validation steps are needed to demonstrate that an identified biomarker is a reliable predictor, and also that the process of converting such a biomarker into a practical clinical test is even more daunting (p. 1053, see the paragraph bridging the left and right columns). Baker ("In Biomarkers We Trust?" Nature Biotechnology 2005; 23(3),297-304) also speaks to the unpredictability involved in clinically applying biomarkers (see p. 298, the section "Walking on Thin Ice"): "Using a new biomarker is like walking across a frozen lake without knowing how thick the ice is," says Ole Vesterqvist ..."You start walking, and you get comfortable. Then you break through." The Examiner concluded that "the state of the art teaches the unpredictability associated with the clinical use of biomarkers even after a biomarker has been correlated with a specific disease state".

In the Barasch/Devarajan 132 Declaration at paragraph 20, the Applicants explain that a successful or "ideal" diagnostic biomarker would be characterized by three features: a) its expression would be tightly correlated with the disease; b) it would be easily (non-invasively) detectable, and c) it would allow detection at the earliest stage possible. The question to be addressed by the Examiner is therefore whether the skilled person, with Matthaeus in hand, **would** have identified NGAL as a biomarker of renal ischemia. However, in fact, the kidney NGAL described by Matthaeus is present at i) the wrong place, ii) at the wrong time, and iii) in the wrong chemical configuration.

Obviousness requires that there was *a reasonable expectation of success*. The relevant question is whether it was obvious for the skilled person to try the approach with a reasonable expectation of success, which should not be confused with the understandable "hope to succeed". what matters is whether "...*this* [skilled] *person would have any reasonable expectation of success when embarking on it* [= an experiment]." In other words, what lastly counts is whether the skilled person -relying on his expert knowledge of the field at issue- would have objectively (i.e. without logical flaw and in a verifiable manner) concluded that an approach (or experiment) **would** have worked.

Barasch/Devarajan 132 Declaration at paragraph 21 notes that feature a) of a diagnostic biomarker requires that the expression of the biomarker in the sampled medium (e.g., urine) be tightly correlated with the disease, in this case ischemic renal injury, including an ischemic renal injury that can progress to acute renal failure.

This is however *prima facie* **not** the case here. Matthaeus identify that NGAL protein is associated with MMP-9 in the injured kidney tissue, and is upregulated in the injured proximal renal tubuli of the ischemic kidney, but do not mention or suggest the urine. Consequently, Matthaeus teaches nothing about a correlation of the expression of urinary NGAL with ischemic renal injury, let alone "a tightly correlated expression" with a renal tubular cell injury. Matthaeus provide only a single datum on the expression of NGAL, in the proximal tubules, at a time 24 hours after the induced renal injury. Since a "correlation" requires a determination of the strength and direction of a relationship between two variables, the limited data (datum) provided by Matthaeus fails objectively, and subjectively, to establish a "tight correlation" of the expression of NGAL with an ischemic renal injury.

At page 8 in paragraph 8(b) of Dr. Keirans' 132 Declaration, he states that "the Office Action takes as admitted prior art that disease processes may produce changes in the levels of certain specific analytes, and that measurement of the levels of such analytes can be used to detect the presence of the disease. Based on this, the Office Action asserts that it would have been obvious to one of ordinary skill in the art to detect NGAL for the purpose of diagnosing acute renal injury in light of the teachings of Matthaeus 1 or Matthaeus 2 that NGAL is specifically elevated in this disease condition, and further in view of the general knowledge of one skilled in the art that markers changed in response to disease can be used as biomarkers for diagnosis of the disease. However, these teachings, either alone or in combination with any of

the other applied references, miss a logical connection between the art of record, and the invention as supplied by the subject application. Namely, ***even if*** the teachings of the prior art are accepted as characterized in the Office Action, for NGAL to function as biomarker allowing a viable NGAL assay to be obtained there is a need, amongst other things, for (a) specificity of the target molecule being assayed for the disease at issue, (b) specificity of the target molecule for the sample type being assessed, and (c) sensitivity of the assay for the target molecule being detected. For all the reasons discussed in the paragraphs below, this need was not satisfied by the prior art, but only by the applicants' invention."

It is clear from the Matthaeus et al. references that the authors themselves, as would a person of ordinary skill in the art, had no expectation of NGAL as a urinary biomarker of ischemic renal injury at the time of the present invention.

(f) sampling of NGAL in urine is not predictable

Applicants point out that, only after the Examiner made the inaccurate conclusion that Matthaeus et al. would lead a person of ordinary skill to the conclusion that NGAL has the properties and attributes of a biomarker, did the rejection then further extend – improperly - the teaching of Matthaeus et al. beyond its actual limited description, based solely on this alleged obviousness of NGAL as a biomarker.

Having erroneously established (page 8 lines 14-17) that "one of ordinary skill in the art at the time of the invention (would) conclude from the finding of Matthaeus 1 or 2 that NGAL *could be* used as a biomarker of renal ischemia," (emphasis added) the rejection goes on to state "it would have been further obvious to employ urine as the sample source, rather than the kidney tissue samples examined in the rat models of Matthaeus 1 and 2, for the following reasons (emphasis added)". The erroneously-based "further obviousness" finding by the rejection concludes on the next page that "(o)ne skilled in the art would immediately recognize that isolation of kidney tissue would be very invasive and therefore unsuitable method for diagnosing renal injury in humans," and that "in light of the general knowledge of one skilled in the art that urine is an easily collected and non-invasive sample source for assay of biological analytes (as taught for example by Ramsden et al.), it would have been obvious to use urine as the sample

source instead of kidney tissue samples when detecting NGAL for diagnosis of ischemic renal injury in human subjects, for the advantage of being a non-invasive and easily collected sample."

Applicants traverse these statements and conclusion.

Applicants do not agree with this fact finding of Ramsden et al, and note that the rejection points to Ramsden et al. as teaching that urine samples are noninvasive and convenient, but such teaching is so general as to be lacking of any factual finding whatsoever. Applicants state their opinion in the Barasch/Devarajan 132 Declaration at paragraphs 24 and 26 that the authors of Matthaeus had no expectation of NGAL as a urinary biomarker of ischemic renal injury, and a person of ordinary skill in the art reading Matthaeus would not have considered, and could not have predicted, that NGAL would be a urinary biomarker of ischemic renal injury at the time of the present invention. Ramsden relates to detection of the marijuana-metabolite THC as a drug marker in urine, and makes no mention or suggestion of biomarkers or renal injury or disease.

On the contrary, Applicants point out at paragraph 27 of the Barasch/Devarajan 132 Declaration that previous responses in the examination of this application that asserted that a person of ordinary skill would have found that isolating kidney tissue (either excising portions or homogenizing the whole kidney) was entirely necessary and appropriate for the purposes of study that was the subject of Matthaeus, which was to observe the pattern and location of mRNA and protein expression in a post-ischemic kidney. The invasiveness of a method depends on the context of its use. For the purpose identified in Matthaeus of studying the synthesis and co-expression of NGAL, MMP-9 and TIMP-1, Applicants assert that a person of ordinary skill would find the invasive isolation of kidney tissue entirely appropriate (see our remarks in the Response of June 3, 2008, pages 15-16). The stated findings of Matthaeus 1 and 2, that NGAL protein expression was upregulated after 24 and 48 hours, and that immunocytochemistry of NGAL revealed bright fluorescence in the most extensively damaged areas (the "injured proximal tubuli") could not have been determined if urine sampling were substituted for the methodology of Matthaeus. Therefore, for the purposes taught in Matthaeus 1, a person of ordinary skill in the art would consider urine sampling neither a suitable nor obvious substitute for the method of isolating kidney tissue disclosed in Matthaeus.

Further, Dr. Keirans states in at paragraph 8(c) of his 132 Declaration that the overwhelming majority of diagnostic tests that measure proteins use blood as the matrix. Urine

is rarely the diagnostic media of choice. The problem with urine is that even if a protein is observed in the urine (whether made by the tubules or present during filtration), the impact of absorption by the tubules en route to making urine can make the urine concentration of the protein diagnostically useless. In other words, the mere presence of NGAL in the urine does not make it useful. Furthermore, as discussed in 9 below, absent the applicants' invention, there was no apparent specificity of NGAL for either renal disease - - much less for renal tubular cell injury - - or for urine. It should be emphasized that while the Office Action relies on Moses et al. to support use of urine as an appropriate biological specimen in assessing NGAL, the reference more prominently discloses both free forms and complexes of NGAL with MMP-9 in urine – and the conclusion that this provides evidence for NGAL modulating MMP-9 activity in cancer patients. MMP-9 is known to directly play a role in remodeling and repair functions (Moses et al.) and Matthaeus et al. state MMP-9 and NGAL can form covalent disulfide complexes. An alternative conclusion from the combined references is that MMP-9 is serving it's known role of remodeling and repair functions in acute ischemic renal failure, and that upregulation and expression of NGAL is a feedback mechanism imposed as a regulatory function of NGAL on MMP-9 to prevent it's in vivo degradation. Experimental designs in the cited references are incapable of isolating and defining the role of NGAL in acute kidney injury that is identified in the instant invention.

(g) lack of predictability of success

Feature b) of the diagnostic biomarker requires the possibility of easy detection. This feature is not disclosed or suggested by Matthaeus. Matthaeus do not indicate an alternative "window of observation" for NGAL that would allow instead for its non-invasive monitoring. When it comes to **where** the NGAL actually shows up, Matthaeus do not go beyond saying that NGAL protein expression was "upregulated in the injured proximal tubuli."

In conclusion, the rejection first has employed a series of short skips involving alleged "common knowledge of persons skilled in the art" to make the greater leap, from the plain teaching of Matthaeus, to a factually unsupportable position that "it would have been obvious to one of ordinary skill in the art at the time of the instant invention to conclude from Matthaeus that NGAL could be used as a biomarker of renal ischemia, such that it would have been obvious

to detect NGAL in human subjects for the clear purpose of diagnosing human disease." Matthaeus neither established an objectively "tight correlation" of the expression of NGAL with the ischemic renal injury, nor provided any window of observation of the expression of NGAL that can be easily detected. This is because the observed proximal renal tubuli-related NGAL of Matthaeus (despite its association with MMP-9) must be able to pass from the proximal renal tubular cells into the urine, something which a person of ordinary skill would clearly not predict or considered.

(h) differences between Matthaeus and the claimed invention

The rejection at page 7 lines 14019 says that Matthaeus differs from the claimed invention by failing to specifically teach detecting NGAL in urine, by failing to teach detecting NGAL within four hours, and by failing to detect NGAL as an antibody-NGAL complex. While certainly true, the findings of fact also make clear that Matthaeus differs from the claimed invention in failing to teach detecting of a renal tubular cell injury (since it is not certain from Matthaeus that the appearance of NGAL in the proximal tubuli *was* detecting renal tubular cell injury), and in failing to teach that the level of NGAL, in the urine or otherwise, correlates with the mammal having the renal tubular cell injury. Further, the "general knowledge" of a person of ordinary skill in the art would not have made these differences non-obvious or predictable.

(i) lack of reasonable expectation of success - secondary references

The rejection goes on at page 9 line 14 to page 10 line 2 to state that "(o)ne would have a reasonable expectation of success because it was known in the prior art that NGAL is excreted in urine, as taught by Blaser et al. and Moses et al. (US 7153660). In particular, Blaser et al. teach detection of human neutrophil lipocalin (NGAL) in urine by sandwich ELISA (see in particular the abstract; page 139, section 2.4; and pages 142-143, sections 3.3-3.4). Moses et al. also teach that NGAL may be detected in human urine by Western Blot (Example 2 and Figure 1B). As such, in light of the teachings of Blaser et al. and Moses et al., one skilled in the art would have a reasonable expectation of success in using urine as a sample source for detection of NGAL in response to renal injury (rather than kidney tissue as taught by Matthaeus 1 and 2) since NGAL was known at the time of the instant invention to be excreted in urine."

Applicants traverse such conclusion.

As stated in Barasch/Devarajan 132 Declaration at paragraph 41, Applicants do not agree with this characterization of Moses and Blaser, none of which is reasonably related to ischemic renal injury or other renal tubular cell injuries, and none of which identify NGAL specifically as a diagnostic biomarker of any specific disease. Applicants note while both Moses and Blaser teach one is capable of detecting NGAL in a sample of urine, neither reference provides any support for any reasonable expectation of success that NGAL would be identified in the urine of a patient following an ischemic renal injury, if only because these references addressed work on different diseases.

The Barasch/Devarajan 132 Declaration at paragraph 42-45 describes that Moses (US 7,153,660) relates to a method for diagnosing the presence of or prognosing appearance of metastatic cancer, by the identification of high molecular weight enzyme complexes comprising MMPs in bodily samples (abstract). The cancer is identified as organ-confined prostate cancer, metastatic prostate cancer, cancer found in cells of epithelial origin, mesodermal origin, endodermal origin or hematopoietic origin, and cancer selected from the group consisting of cancers of the nervous system, breast, retina, lung, skin, kidney, liver, pancreas, genito-urinary tract, and gastrointestinal tract. (column 2 lines 22-29). The high molecular weight enzyme complexes comprising can also comprise a lipocalin, e.g., NGAL, and/or a TIMP, e.g., TIMP-1 (column 2 lines 35-37). The method also includes obtaining a biological sample from a subject and detecting lipocalin in the biological sample, and correlating the presence or absence of the lipocalin with the presence or absence of a tissue remodelling-associated condition, thereby facilitating the diagnosis of the subject for a tissue remodelling-associated condition (column 2 lines 48-54).

Most of the details of the Moses reference relate to supporting the finding that the 125 kDa MMP activity in urine samples of cancer patients was a complex of MMP-9 and NGAL (column 15 lines 37-39). Moses states that the origin of the 125 kDa MMP-9/NGAL activity in urine of cancer patients remains unclear, but that it is likely that MMP-9 and NGAL are separately executed into urine where they form the 125 kDa MMP-9/NGAL complex. (column 17 lines 37-48). At column 5 lines 22-38, Moses states that "(m)any thousands of proteases occur naturally, and each may appear at different times of development and in different locations in an organism. The invention herein features enzymes of the class of the matrix

metalloproteinases (MMPs, class EC 3.4.24). These enzymes, which require a divalent cation for activity, are normally expressed early in the development of the embryo, for example, during hatching of an zygote from the zona pellucida, and again during the process of attachment of the developing embryo to the inside of the uterine wall. Enzyme activities such as N-acetylglucosaminidase (EC 3.2.1.50) appear in urine in the case of renal tubular damage, for example, due to diabetes (Carr, M. (1994) J. Urol. 151(2):442-445; Jones, A, et. al. (1995) Annals. Clin. Biochem., 32:68-62). That these activities appear in urine as a result of renal tubular damage is irrelevant to the *present invention as described herein." (emphasis added)*

Therefore, the only mention in Moses et al. of a renal disease, other than kidney cancer as one of numerous metastatic cancers, is a reference to "renal tubular damage" associated the enzyme activity of N-acetylglucosaminidase in the urine, which activity is "irrelevant to the present invention" of Moses et al. This would suggest to us an absence of any correlation or association between the diagnosis of a metastatic cancer by detection of the MMP-9/NGAL complex in urine, and renal tubular damage, and does not predict any success in identifying NGAL in the urine of a patient following an ischemic renal injury.

We also note that Moses et al. relates exclusively to a non-acute disease, specifically metastatic cancer, and that it is not known which latency period may expire between the development of such cancer and the appearance of NGAL in the urine.

The Barasch/Devarajan 132 Declaration at paragraph 46 describes that Blaser relates to detecting the level of NGAL (8 μg/L = 8 ng/mL) in pooled urine samples in healthy donors over a 24 hour period. (See the last sentence in section 2.4 Preparation of the samples, on page 139). Thus, this reference implies only that some level of NGAL is found in the urine of alleged "healthy" persons. Blaser does not report or suggest that the kidney is a source of the observed NGAL, or has even accessed the urine through the kidneys. Indeed, the source of NGAL in the urine of Blaser may be completely unrelated to the kidney (such as, the bladder). It is well known that the urine contains very few proteins (dozens perhaps), compared to the thousands of known proteins expressed within the tissues of the kidney itself, and that source of a protein appearing in the urine cannot easily be predicted as having been sourced from the kidney, as opposed to the bladder, prostate (in men), vaginal tissue (in women), blood, or other source. Since the site of origin of urinary NGAL from Blaser is simply not known, Blaser's association with Matthaeus is unsupportable, and - at best - improper hind-sight.

The Office Action at page 10 lines 11-17 goes on to state that "Ohlsson et al. found that greatly elevated NGAL levels are strongly correlated with decreased renal function (emphasis in the rejection)", and that "(t)aken together with the finding of Matthaeus 1 or 2, it would have been obvious to detect NGAL for the purpose of diagnosing renal dysfunction since the reference established that NGAL is specifically elevated in this disease condition, and further in view of the general knowledge of one skilled in the art at the time of the invention that markers changed in response to disease can be used as biomarkers for diagnosis of the disease."

Applicants traverse this characterization of Ohlsson et al.

As stated earlier, the examiner's alleged "common knowledge statement" that the general knowledge of one skilled in the art at the time of the invention includes that markers changed in response to disease can be used as biomarkers for diagnosis of the disease, fails because it is at best a generalized principle that is devoid of specifics. The alleged common knowledge statement actually was "disease processes generally may produce changes in the levels of certain specific analytes, and that measurement of the levels of such analytes can be used to detect the presence of the disease". This asserted "common knowledge statement" lacks the specifics for any person to either support or refute the statement, and was understood by Applicants to be a generalized concept that may or may not be true depending on the specifics, which are completely lacking. That is, the alleged statement may or may not be true, and unpredictably so, depending on the specific disease, the specific analyte, and the timing and degree of change in the level of specific analyte. And any failure by Applicant to have refuted such statement does not extend the scope of the statement to any specific disease or analyte in any way.

The Barasch/Devarajan 132 Declaration at paragraph 48-52 describes that Ohlsson et al. states specifically that "greatly elevated NGAL levels were seen in our patients. However, there was a strong correlation with decreased renal function" (page 531, the left column, last paragraph). The exact phrasing of the statement made in Ohlsson et al. is important. Ohlsson et al. made a general statement that their patients had elevated levels of NGAL. When the authors noted that the NGAL level correlated with elevated levels of cystatin C (the renal function markers used by the authors in the study), they "corrected" the NGAL result dividing the NGAL level by the cystatin C level, to normalize the result with "renal function". It is clear that Ohlsson et al. taught that cystatin C was the renal function marker, and that in these particular

patients, the level of NGAL showed a correlation with renal function. This was not to convey that the level of NGAL correlates with renal function generally.

In the Barasch/Devarajan 132 Declaration at paragraph 49, Applicants refer to the "Ohlsson Declaration" submitted by Dr. Prasad Devarajan in the co-pending application 11/096,113, which was submitted with the response and the RCE filed on February 20, 2007, related to the Ohlsson et al. reference. A copy of such 132 Declaration is attached to the Barasch/Devarajan 132 Declaration as Appendix A.

The Ohlsson Declaration mentioned above stated Dr. Devarajan's opinion that the authors of Ohlsson et al. (2003) clearly attribute the result (an elevated NGAL levels that correlated with decreased renal function) to neutrophil activity caused by some factor other than renal dysfunction, for which cystatin C was their marker, since the authors normalize the NGAL level to cystatin C, and they attribute the increase in neutrophil degranulation to the treatment of the DC group with immunosuppressive medication. (It is well known that neutrophils are a major source of MGAL in the blood, the sampling source in Ohlsson et al).

Applicants also noted that Ohlsson et al. describes "renal dysfunction" as opposed to "renal injury". Since it is well known to person of ordinary skill in the art that a downturn in renal function trails substantially an earlier, undetected kidney injury (typically by a couple of days), even the correlation of the level of NGAL with kidney dysfunction is not predictive of a biomarker of the early onset of a renal tubular cell injury.

Applicants also state their belief at paragraph 52 that the detection of the level of NGAL in a serum sample would have revealed nothing to a person of ordinary skill concerning the level of NGAL in urine, not unlike the skilled person's understanding that increases in the level of creatinine in the serum are not predictive of an increase in the level of creatinine in urine.

Finally, Applicants also note that the Office Action refers to David et al. (US 4,376,110), which relates to now well known ELISA techniques. David makes no mention of renal injury or disease, or its detection via the urine, or even the "renal", "kidney", "disease", 'injury" or "urine". David et al. does teach that the technique reduced significantly the time for running the assay.

(j) Matthaeus and other references fail to predict the early onset of a urinary biomarker

The rejection at page 12 lines 4-9 notes "that Matthaeus 1 and Matthaeus 2 each teach that NGAL was elevated in kidney (tissue) "after 24 and 48 hours" of renal injury (ischemia induced by operation). Matthaeus 1 also clearly teaches NGAL upregulation in the context of *acute* ischemic renal failure (title). However, the references do not discuss whether samples were also taken at other, earlier time points." The rejection emphasizes the word "acute" in an apparent attempt to suggest that some motivation might attach to look for NGAL at some time earlier than 24 hours.

Applicants traverse.

Without recourse to Applicants' own teachings, it would not have been reasonable for a person of ordinary skill in the art to consider that the authors of Matthaeus 1 or 2 ever had any time earlier than 24 hours in mind; there is no motivation or suggestion in Matthaeus that they were ever thinking about biomarkers.

Rather, contrary to the rejection, the combination of Matthaeus 1 or 2, alternatively with Ohlsson et al), in view of Ramsden, Moses, Blaser and David, fails to identify that NGAL is predictably a urinary biomarker for renal tubular cell injury; that is, a detectable protein tightly correlated with the disease in a non-invasively sampled body fluid. Matthaeus 1 and 2 describe NGAL late-term upregulation of NGAL with MMP-9 in the injured proximal tubules following an ischemic renal injury, but lack any predictability of the presence of NGAL in the urine at the early onset of the ischemic renal injury. Ohlsson et al. relates to a correlation of plasma NGAL with a renal function biomarker (cystatin C) months, if not years, after any suspected event that might have been the onset of any renal injury or dysfunction. Ramsden teaches detection of the marijuana-metabolite THC as a drug marker in urine. Moses relates to detection of an NGAL/MMP-9 complex in the urine of metastatic cancer patients; Blaser describes urine levels in 24-hour pooled urine samples from healthy subjects. David teaches an ELISA *per se*.

Consequently, new Claims 67-84 are patentable over the rejection.

(k) Muramatsu et al. does not establish *prima facie* obviousness

The rejection also relies on Muramatsu et al. to support the *prima facie* rejection as teaching that detecting urinary Cyr-61 protein within 3-6 hours, 6-9 hours, 9-12 hours, 12-18 hours and 18-24 hours after ischemia. More specifically, Muramatsu et al. teach detecting the

presence of Cyr61 protein extracted out of mouse urine onto heparin beads, to detect an ischemic renal injury. Muramatsu et al. makes no reference to NGAL.

Applicants traverse.

First, Applicants at paragraph 54 of the Barasch/Devarajan 132 Declaration assert that a person of ordinary skill in the art would not, in view of Matthaeus 1 and 2, Ohlsson, Ramsden, Blaser, Moses, and David, have any reason to predict that urinary NGAL is a biomarker of renal ischemia, let alone one that appears within 24 hours in the urine. First, the rejection lacks any reasonable rationale of a suggestion to combine the Muramatsu et al. reference with Matthaeus or any of the other secondary references of Ramsden, Blaser, Moses and David. The predictability alleged by the rejection by further considering Muramatsu et al, which detects a completely different protein within several hours of renal ischemia, is completely lacking. The rejection provides no rationale why a person of ordinary skill having Matthaeus in hand, would ever consider the characteristics of urinary Cyr-61 and the work of Muramatsu *per se*, let alone predict that these might be found in NGAL, certainly not without knowledge of Applicants' invention.

The rejection greatly exaggerates any association that a person of ordinary skill in the art might possibly make between urinary Cyr61 in Muramatsu, and the detection of NGAL and MMP-9 in the injured proximal tubuli in kidney tissue of Matthaeus after 24 and 48 hours. Other than a rat/mouse model of ischemic renal injury (although Matthaeus do not describe with any specificity the mode of inducing ischemia), there are substantial differences between the Muramatsu and Matthaeus references. The respective authors of Muramatsu et al. and Matthaeus et al. were looking at different proteins (Cyr-61 versus NGAL and MMP-9), in different locations (in urine, versus in the kidney), at different times (within hours, versus after 24 hours), and for different reasons (detecting ischemic injury, versus detecting the kidney's response to injury). Other than the "who" (the mouse), the "what", "when", "where" and "why" between Muramatsu and Matthaeus are different.

(I) Ohlsson et al. and Muramatsu et al. are not citable as prior art

Notwithstanding the traversal of the rejections based upon Ohlsson et al. and Muramatsu, the rejection relying upon Ohlsson et al. and Muramatsu also fails to state a *prima facie* obviousness because these two references are not prior art against Applicants' claims.

Declarations under 37 CFR 1.131 used to antedate a referenced are not required to show the invention "defined in the claim the applicant is asking for" (*In re Stempel;* 113 USPQ 77, 78). To antedate a reference, including a reference in a combination of references in an obviousness rejection under 35 U.S.C. 103(a), the 131 declaration needs only to show (a) what the antedated reference shows, and (b) possession of either the whole invention claimed or something falling within the claim, in the sense that the claim as a whole reads on that something (see *In re Tanczyn*, 146 USPQ 298, 301, emphasis added). "One of ordinary skill in the art would be satisfied from the facts shown in the affidavit that (Applicants) had completed *the invention* as defined in the claims" (quoting from *In re Spiller,* 182 USPQ 614, emphasis in the original), even though the Supplemental 131 Declaration may be alleged to not disclose every detail of the claimed invention.

Muramatsu et al. is relied upon in the 103(a) rejection of the Office Action to show "screening for a biomarker of ARF (Cyr61) by detecting the presence of urinary Cyr61 within specified times in relation to the onset of induced renal ischemic, as a model of ARF" (see especially pages 1603-1604, "Urine Collection", page 1606, and figure 8). The referenced exemplifies time points of 3-6, 6-9, 9-12, 12-18, and 18-24 hours after ischemia (see especially the legend to Figure 8)" (pages 19-36 of the Office Action mailed 09/04/2008).

The quoted pages in Muramatsu et al. described the collection of urine and analysis of the quantities Cyr61 in the urine by SDS-PAGE and western blotting, following 30 minute bilateral ischemia induced by clamping the renal arteries of a rat.

It is clear that the Applicants' Supplemental 131 Declaration shows for NGAL everything that Muramatsu et al. shows for Cyr61 and that for the respective species of the renal biomarkers, the Supplemental 131 Declaration shows that the Applicants have priority with respect to so much of the claimed invention as the Muramatsu et al. reference happens to show. Muramatsu et al. is cited to establish the detection of a biomarker of ARF (genus) by disclosure of a species (Cyr61). Applicants' earlier possession of the different species NGAL from the same genus therefore antedates Muramatsu et al. with respect to Claim 66, as well as dependent claims thereto, and new claims 67-84.

Furthermore, the 131 Declaration also shows for NGAL in a body fluid sample species (urine) of a mammalian subject species (a mouse) following a renal tubular cell injury (an ischemic renal injury) everything that Ohlsson et al. is alleged to show for NGAL in a different

body fluid sample species (plasma) of a different mammalian species (a human) following what at best might be described as a renal injury secondary to ANCA-associated systemic vasculitis, and that Applicants' earlier possession of NGAL in urine antedates Ohlsson et al.

It is also clear that the Applicants' Supplemental 131 Declaration demonstrates Applicants' possession of "something falling within the claim", in the sense that the claim (Claim 1 or Claim 30 pending at the time of the 09/04/2008 Office Action, and Claims 66, 67, 68 and 82 presently pending) reads on the demonstrated showing in Applicants' Supplemental 131 Declaration. The differences which may exist between the claimed invention and the specific reduction to practice established by the Supplemental 131 Declaration showing are such as would be obvious to one of ordinary skill in this art. (See *In re Spiller*, supra at 620).

Notwithstanding the rejection's failure to find that the 131 Declaration has antedated both Muramatsu et al. and Ohlsson et al. on the basis of showing what the antedated references show, and (b) possessing either the whole invention claimed or something falling within the claim, Applicants further assert that the submitted 131 Declaration executed by joint inventors Prasad Devarajan and Jon Barasch also established that the invention as claimed was conceived and reduced to practice at a time that pre-dates the effective reference date of Muramatsu et al. and Ohlsson et al. (February 28, 2003).

With respect to claim 66 and claims depending therefrom (except claims 37 and 60), the rejection at page 31 asserts that the experiments described in the 131 Declaration are not commensurate with the scope of the claims, because the claims are diagnostic of a present injury, while the experiments induced the injury surgically and therefore knew the injury would be present beforehand.

Applicants traverse. With all due respect, experimentation to determine the effectiveness of an invention often requires that the condition that one intends to detect be induced so that the means of detecting can be correlated with the condition. The experiments unquestionably detected the injury to the kidney, induced or otherwise.

Finally, the Examiner states that "(n)o statements regarding diligence have been advanced by Applicant(s)". Applicants asserted in the 131 Declaration that the experiment was a reduction to practice of the claimed invention, which include the presently amended and new

claims. Applicants request that the Examiner reconsider the need for diligence in such circumstance.

With respect to claims 37 and 60, which were denied priority to the provisional filing dates, the Muramatsu et al. and Ohlsson et al. references are deemed 102(b)-type references, which cannot be antedated under 37 CFR 1.131. Nonetheless, Applicants note that the rationale for the denial of priority is the lack of an express disclosure in the provisional applications for the events "open heart surgery" and "cardiac surgery". Applicants note that, on such narrow scope, neither does Muramatsu et al. or Ohlsson et al. disclose expressly or inherently "open heart surgery" and "cardiac surgery".

Claims 4-5, 9-11, 33, 35, 55, and 66 are rejected under 35 U.S.C. 103(a) as being unpatentable over either "Matthaeus 1" or "Matthaeus 2", in view of "Ramsden", "Blaser", "Moses", "David", and further in view of Sanicola-Nadel et al. (US 6,664,385).

Applicants traverse.

Applicant incorporates by reference here the arguments above with respect to the teachings and fact findings, and arguments pertaining thereto, of either "Matthaeus 1" or "Matthaeus 2", in view of "Ramsden", "Blaser", "Moses", "David". Applicants also assert that new Claims 67-84 are patentable over the rejection.

Sanicola-Nadel et al. relates to the KIM-1 protein. NGAL and other urinary proteins are not disclosed or suggested.

The rejections states at page 16 lines 5-6 that Sanicola-Nadel teaches use of KIM-1 in a diagnostic method, in the abstract and at col 1 lines 49-55. For the record, the only mention of KIM-1 protein and diagnostic methods is at col 2 line 66 to col 3 line 21, and at col 17 lines 34-41. The rejection alleges that Sanicola-Nadel et al. teaches that KIM-1 protein is selectively upregulated at any time within one week following any insult that results in injury to kidney tissue, mentioned at col 5 lines 17-27.

Applicants assert that the rejection formulates an inaccurate finding of facts in Sanicola-Nadel.

Applicants clarify and correct the characterization made by the examiner, at paragraph 57 of the Barasch/Devarajan 132 Declaration, that Sanicola-Nadel at col 5 lines 27 refers to the time of the selective upregulation of *KIM-1* mRNA, and not to the expression or appearance of KIM-1

protein *per se*. Moreover, a person of ordinary skill in the art would understand that Sanicola-Nadel speculates, rather than teaches, that such upregulation of mRNA "might be identified" at 10 hours. While such time is "within one week", the time course for appearance of KIM-1 protein within the urine or serum is not addressed at all.

The Applicants also note (paragraph 58) that the detection of KIM-1 protein is from a homogenized kidney that is excised 24 hours and 48 hours after unilateral kidney ischemia (see column 17 line 66 to column 18 line 8). Specifically, kidney homogenates of *contralateral and post-ischemic kidneys* were examined following a 40 minute clamping *of the renal artery and vein of a single kidney* for each rat. It was well known to persons skilled in the art that unilateral renal ischemia does not necessarily result in the development of acute renal failure, which we also expressly teach in the instant application (our paragraph [0085]). Traditional markers of ischemic renal injury, including serum creatinine, are not usually elevated in a unilateral ischemic rat or mouse model. Therefore, a person of ordinary skill in the art would understand that the authors speculate that KIM-1 might be found in either the urine or the serum of the disclosed rat models of Sanicola-Nadel, since no urinary or serum renal marker was known at the time of Sanicola-Nadel or at the time of the present invention, had been shown to appear within 24 hours of an ischemic renal injury, and no actual showing of KIM-1 in urine or serum is actually shown.

The Applicants also state at paragraph 59 of the Barasch/Devarajan 132 Declaration that they would not predict that the results observed for KIM-1 protein would be applicable to NGAL for numerous reasons. KIM-1 protein is quite different from NGAL in properties and size; for example, NGAL is a secreted whole protein, while in the case of KIM-1, only a small ectodomain of the protein is proteolytically processed and appears in the urine. That is to say, any appearance of the ectodomain of the KIM-1 protein in urine involves a kinetically-driven, time-consuming biological process, with an unpredictable duration before its appearance in the urine. NGAL at the time of the present invention was generally associated with the degranulation of activated neutrophils. Any prediction or suggestion based upon KIM-1 protein could not carry over to NGAL, and *visa versa*.

Claim 2 is rejected under 35 U.S.C. 103(a) as being unpatentable over either "Matthaeus 1" or "Matthaeus 2", in view of "Ramsden", "Blaser", "Moses", "David", and

"Muramatsu"; or, in the alternative, over either "Matthaeus 1" or "Matthaeus 2" and "Ohlsson", in view of "Ramsden", "Blaser", "Moses", "David", and "Muramatsu"; or over either "Matthaeus 1" or "Matthaeus 2", in view of "Ramsden", "Blaser", "Moses", "David", and further in view of Sanicola-Nadel et al; further in view of Valkirs et al. (US 2003/0109420) or Linzer et al. (US3,635,091).

Without acquiescing to the above rejection under 35 USC 103(a) or its basis or rationale, Applicants request reconsideration and withdrawal of the rejection in view of the amendments to the claims from which Claim 2 depends.

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Claim 55 is rejected under 35 U.S.C. 103(a) as being unpatentable over either "Matthaeus 1" or "Matthaeus 2", in view of "Ramsden", "Blaser", "Moses", "David", and "Muramatsu"; or, in the alternative, over either "Matthaeus 1" or "Matthaeus 2" and "Ohlsson", in view of "Ramsden", "Blaser", "Moses", "David", and "Muramatsu"; or over either "Matthaeus 1" or "Matthaeus 2", in view of "Ramsden", "Blaser", "Moses", "David", and further in view of Sanicola-Nadel et al; further in view of Kosako (US 5,527,714).

Without acquiescing to the above rejection under 35 USC 103(a) or its basis or rationale, Applicants request reconsideration and withdrawal of the rejection in view of the amendments to the claims from which Claim 55 depends.

Claim 60 is rejected under 35 U.S.C. 103(a) as being unpatentable over either "Matthaeus 1" or "Matthaeus 2", in view of "Ramsden", "Blaser", "Moses", "David", and "Muramatsu"; or, in the alternative, over either "Matthaeus 1" or "Matthaeus 2" and "Ohlsson", in view of "Ramsden", "Blaser", "Moses", "David", and "Muramatsu"; or over either "Matthaeus 1" or "Matthaeus 2", in view of "Ramsden", "Blaser", "Moses", "David", and further in view of Sanicola-Nadel et al; further in view of Zanardo et al.

Applicants traverse.

Applicants traverse the rejection's construction of Zanardo et al, which therefore overcomes the alleged *prima facie* obviousness basis of the rejection. Specifically, Zanardo et al. cannot be found to have stated that its authors were assessing "renal injury", as alleged. Rather, as Zanardo et al. expressly states, they used serum creatinine to assess "renal function".

As a person of ordinary skill would readily understand, renal injury and renal function are not synonymous.

Furthermore, in the action the examiner states "with respect to the recitation (in the claim) that the sample is obtained at 2 hours,... Zanardo et al. measured serum creatinine as a marker of renal function at the time of weaning from CPB (see Table II). As listed in the Table, bypass time was approximately 2-3 hours (see "CPB time (min)").

The Applicants assert that the Examiner specifically state exactly *where* these features are being taught by Zanardo et al, and also explain why any such alleged sampling and measurement of serum creatinine is nothing more than a baseline, as opposed to one for the purpose of measuring a change in renal function resulting from CPB, which is typically understood to occur, if at all, at 24-48 hours post-CPB.

Secondary Evidence of Patentability

Notwithstanding Applicants' traversal of the above 35 USC 103 rejection of the claims under examination, including the new claims added, Applicants present evidence of secondary considerations," such as "... long felt but unsolved needs, [and] failure of others." *Graham v. John Deere Co.*, 383 U.S. at 17, 148 USPQ at 467.

In his 132 Declaration at paragraphs 9(a)-(f), Dr. Keirans presents evidence that the claimed invention provided success for the long felt but unsolved need for a marker of renal tubular cell injury, including ischemic renal injury, that can be detected at less than 24 hours after the onset of the injury, and evidence that failed to identify a urinary biomarker for renal tubular cell injury, and failed to specifically identify NGAL as such.

Dr. Keirans states that prior to the invention of the subject application, there had been relatively little success in identifying biomarkers that could be used at an early stage for an assay for acute renal tubular cell injury. Whereas markers that can assess renal function later than 24 hours of the onset of renal injury and after are available, particularly serum or plasma creatinine, a urine assay for detecting renal tubular cell injury (RTCI) within 24 hours of a suspected renal injury was not. Such an assay of RTCI requires a biomarker that (a) increases or changes in value at less than 24 hours, (b) is specific for the sample type being assessed (i.e., urine), (c) is specific for disease (i.e., renal disease) or preferably source of disease (i.e., renal injury) being assessed, and (d) is sensitive for the target molecule (i.e., NGAL) being detected.

In the context of a renal assay, finding a biomarker that increases in value at within less than 24 hours of a suspected RTCI is particularly difficult. Prior to the invention of the subject application, others had tried to identify such a marker (see, e.g., Muramatsu et al., cited in the Office Action and published after the Matthaeus et al. references). Cyr61 was a secretory protein demonstrated by Muramatsu et al. to be upregulated in rat ischemia model and secreted into urine using RDA, northern and southern blots and immunohistochemistry (essentially same experimental approaches as Matthaeus et al.). Cyr61 was analyzed at 42 kD and it was noted that several non-specific higher molecular weight bands were also found. Although a specific function for Cyr61 was not determined, it was present in urine within 3-6 hours and peaked at 6-9 hours following renal injury. It is surprising that if detecting NGAL in urine rapidly following renal injury was obvious to one of ordinary skill, that Muramatsu et al. failed to identify it in the high molecular weight fraction of urine in their work published after Matthaeus et al. They also did not discuss NGAL nor did they cite Matthaeus et al. in their paper.

Dr. Keirans notes that in view of failure to identify other renal biomarkers that could be effectively applied to assess RTCI within a timeframe of less than 24 hours, at the time of the invention of the subject application, the need for biomarkers for detecting RTCI at less than 24 hours was long-felt, but remained unsolved. An increase in serum or plasma creatinine, detectable at 24 hours or later, was the gold standard for assessing renal function. However, by the time an increase in creatinine can be detected, as much as 50% or more of renal function may be lost. This is because the serum creatinine increase occurs much later than the renal injury resulting in the loss of function.

Dr. Keirans also states that the failures of others to identify biomarkers detectable at within 24 hours of renal injury would lead to general skepticism prior to applicants' invention regarding the use of NGAL to assess acute renal injury. Additionally, there were further more particular reasons at the time of the invention why one of ordinary skill would not consider or recognize urine NGAL for assessing RTCI within 24 hours of suspected renal injury, even in view of the teachings in the prior art. First, NGAL had been detected in tissues and sample types other than urine. NGAL from such other sources may produce a high background, or a complete lack of specificity, precluding use as a marker. At the time of the invention, NGAL mRNA and/or protein had been detected in and associated with neutrophils (see, e.g., Allen et al., Biochimica et Biophysica Acta, 991:123-133 (1989), Kjeldsen et al., Blood 83(6): 1640-1649

(1994), Kjeldsen et al., Blood 82(10): 3183-3191 (1993), and Kjeldsen et al., Blood 83(3): 799-807 (1994)), blood plasma of patients with systemic vasculatis (e.g., Ohlsson et al.) adult bone marrow, uterus, prostate, salivary gland, stomach, appendix, colon, trachea, and lung (e.g., Cowland et al., Genomics 45: 17-23 (1997)), and extracellular fluids of female reproductive tract (Costantini et al., Minerva ginecologica, 54:5, 387-92 (2002 Oct)). Prior to applicants' invention, this detection of NGAL in other than renal tissue or urine would seriously call into question whether NGAL could be assessed in urine as a sample source for kidney injury, distinct from other sources. In particular, appearance of NGAL in blood (red **or** white cells), would raise a question about whether a useful urine NGAL measure could be derived, given the known fact that blood can get into the urine with infection of the kidneys or the bladder, or if there is inflammation due to the presence of stones, immune disorders, allergies, growths anywhere along the genitourinary system, and in many other situations.

Second, at the time of the invention, NGAL had been associated with disease states other than renal disease or RTCI. Venge and colleagues (e.g., Xu et al., Scand. J. Clin. Lab Investigation, 55:125-131 (1995)) determined that serum or plasma NGAL or HNL present in respiratory distress conditions was capable of differentiating viral from bacterial-mediated infection. Moses et al. have documented free and complexed NGAL associated with MMP9 known to be present in several cancers (thyroid, ovarian, breast, colon, etc.). An increase or alteration of NGAL in response to other stimuli would cause one to question whether NGAL could be specifically associated with renal disease or RTCI, particularly within a timeframe of within 24 hours of suspected renal injury. For instance, NGAL had been detected in urine of healthy donors (e.g., Blaser et al.), and increased at the mRNA, and/or protein level with cytokine withdrawal (e.g., Devireddy et al., Science 293: 829-834, (2001)), neutrophil activation, inflammation and infection (e.g., Xu et al., Scand. J. Clin. Lab Investigation, 55:125-131 (1995)); Venge, Allergy, 49(1):1-8 (1994)), cartilage and muscle differentiation (Zerega et al., European journal of cell biology, 79:3, 165-72 (2000 Mar)), and in tumors or cancer (e.g., Bartsch et al., FEBS Lett. 357:255-259 (1995); Gould et al., US 5,627,034; Conklin, US 6,143,720; and Moses et al.). All these associations, particularly with the normal cell, inflammation and infection (which raise issues of potential high background), would call into question prior to applicants' invention whether NGAL could be specifically associated in any way with any renal disease, much less with RTCI. Indeed, upon identification of NGAL in urine

of patients with renal cancer and bladder cancer, Yan et al., J. Biol. Chem., 276:37258-37265 (2001) specifically raise the possibility of neutrophil infiltration as the source of the urine NGAL by stating in the last sentence of the third paragraph in the "Discussion" that: "However, it remains possible, that the urinary MMP-9-NGAL complex may be composed of MMP-9 and NGAL secreted by the neutrophils that have infiltrated the tumor sites."

At paragraph 10, Dr. Keirans states his understanding and belief that one of ordinary skill in the art at the time of the subject invention would not have been able to predict with any certainty based on the art made of record (e.g., Matthaeus 1 and 2) that one would find NGAL in the urine, much less be able to use NGAL as a biomarker in a urine NGAL renal assay. Dr. Keirans also expresses his understanding and belief that one of ordinary skill working in the field at the time of the invention and considering the teachings of the art cited in the Office Action in the context of what all else was happening in the field would have no reasonable expectation that urine NGAL could be successfully employed to assess RTCI at within less than 24 hours of a suspected renal injury.

Based upon the evidence presented above in his 132 Declaration, Dr. Keirans concludes that the findings in the subject application regarding the invention and showing that urine NGAL could be successfully employed to assess renal tubular cell injury (RTCI) at within less than 24 hours of a suspected renal injury were unexpected and not predicted. He notes that this conclusion is confirmed by the guarded optimism set forth in the reference of Stefan Herget-Rosenthal (Lancet, 365, 1205-1206 (2005)), which was published in the same volume as and introduced the studies setting forth the data present in only one of the examples of the subject application.

Finally, Dr. Keirans addresses the success of the present invention, and its exclusive licensing to Abbott. He states that the applicants of the subject application were the first to recognize use of NGAL as a biomarker for assessing RTCI at within less than 24 hours of a suspected renal injury, and that Abbott recognized this upon first learning of the work of Doctors Devarajan and Barasch. Based on Abbott's perception at that time that the results of the inventors now set forth in the subject patent application were truly surprising, and Abbott's perception of the usefulness of NGAL as a renal biomarker, Abbott licensed the intellectual property from the assignees of the Applicants' rights. Dr. Keirans has recommended that Abbott proceed with development and commercialization of the assay to satisfy an unmet diagnostic

need and provide a much needed ability to assess RTCI at within less than 24 hours of a suspected renal injury.

Double Patenting

(i) Co-pending Application No. 11/096,113 (Attorney Docket CHM-025M)

Claims 2, 4-5, 9-11, 33, 35, 37, 55, 60 and 66 are provisionally rejected on the ground of non-statutory obviousness-type double patenting over the claims 2, 4, 7-10 and 22-39 of co-pending Application No. 11/096,113 (Applicants' commonly-owned, co-pending application, Attorney Docket CHM-025M), in view of Ramsden et al., Blaser et al. and Moses et al. Co-pending Application No. 11/096,113 has claims related to detecting NGAL in a sample of blood to identify if the subject is predisposed to progressing to acute renal failure as a result of an acute renal tubular cell injury.

Applicants respectfully request reconsideration and withdrawal of the double patenting rejection, for the following reasons.

MPEP 804 states: "A rejection based on nonstatutory double patenting is based on a judicially created doctrine grounded in public policy so as to prevent the unjustified or improper timewise extension of the right to exclude granted by a patent... and to prevent possible harassment by multiple assignees. Where the claims of an application are not the "same" as those of a first patent, but the grant of a patent with the claims in the application would unjustly extend the rights granted by the first patent, a double patenting rejection under non-statutory grounds is proper.

It is respectfully noted that each and every claim of the present invention requires a "urine sample", while each and every claim of the co-pending application 11/096,113 requires a sample isolated from the blood, including a blood serum sample. Consequently, the claims of these applications cannot be the "same" invention, because there cannot be overlapping of the scope of the claims, and there cannot be any unjustly extending of the rights granted in a first patent issued to one of the applications, by the granting of the second application. The

circumstances described in *In re Schneller*, 397 F.2d 350, 158 USPQ 210 (CCPA 1968), are not applicable in the present circumstances.

Further, Applicants traverse the rejection on the basis that the co-pending claims are drawn to blood or serum, while the instant claims utilize urine. The assertion of obviousness is based solely on Ramsden et al. (that urine is non-invasive), and that NGAL can be detected in urine (Blaser and Moses). Nonetheless, the factual findings pertaining to these references as a whole established earlier in this Response, makes it clear that the rejection fails to establish *prima facie* non-statutory obviousness-type double patenting, because the secondary references utterly fail to teach, suggest or predict that the finding of NGAL in blood or serum following renal tubular cell injury would predictably include the use of urine as a "second window" of detection.

Neither the present application nor the reference co-pending application 11/096,113 have issued as a patent, and the present application has a filing date that precedes the earliest priority date and filing date of the co-pending application 11/096,113.

(ii) Co-pending Application No. 11/770,422 (Attorney Docket CHM-015K)

Claims 2, 4-5, 9-11, 33, 35, 37, 55 and 66 are provisionally rejected on the ground of non-statutory obviousness-type double patenting over the claims 30-31, 33-45 and 47-50 of co-pending Application No. 11/770,422 (Applicants' commonly-owned, co-pending application, Attorney Docket CHM-015K), in view of David et al.

Claim 60 is provisionally rejected on the ground of non-statutory obviousness-type double patenting over the claims 30-31, 33-45 and 47-50 of co-pending Application No. 11/770,422 (commonly-owned, co-pending application, Attorney Docket CHM-015K), in view of David et al, Matthaeus 1 or Matthaeus 2 , and either one of Sanicola-Nadel or Muramatus et al., and further in view of Zanardo et al.

Co-pending Application No. 11/770,422 is a continuation of the present application.

Applicants preserve the right to file a Terminal Disclaimer in the later of the present application and the reference Application No. 11/770,422 that has allowed claims.

(iii) US Application No. 11/770,372 (Attorney Docket CHM-025MK)

Claims 2, 4-5, 9-11, 33, 35, 37, 55 and 66 are provisionally rejected on the ground of non-statutory obviousness-type double patenting over the claims 22, 24-36, 38-42 and 44 of Application No. 11/770,372 (Applicants' commonly-owned, application, Attorney Docket CHM-025MK), in view of David et al., Ramsden et al., Blaser et al, Moses et al, either of Matthaeus 1 or Matthaeus 2 , and either one of Sanicola-Nadel or Muramatus et al.

Claim 60 is provisionally rejected on the ground of non-statutory obviousness-type double patenting over the claims 22, 24-36, 38-42 and 44 of co-pending Application No. 11/770,372 (Applicants' commonly-owned, co-pending application, Attorney Docket CHM-025MC), in view of David et al., Ramsden et al., Blaser et al, Moses et al, either of Matthaeus 1 or Matthaeus 2, and either one of Sanicola-Nadel or Muramatus et al, and further in view of Zanardo et al.

US Application No. 11/770,372 was a continuation of Application No. 11/096,113 (Attorney Docket CHM-025M) discussed above, which is now *abandoned.* Co-pending US Appln. 12/604,117 (Attorney Docket CHM-025MK1C) is a continuation of US Application No. 11/770,372.

Applicants respectfully request reconsideration and withdrawal of the double patenting rejection, with respect to continuation US Appln. 12/604,117, for the reasons stated above.

MPEP 804 states: "A rejection based on nonstatutory double patenting is based on a judicially created doctrine grounded in public policy so as to prevent the unjustified or improper timewise extension of the right to exclude granted by a patent. Where the claims of an application are not the "same" as those of a first patent, but the grant of a patent with the claims in the application would unjustly extend the rights granted by the first patent, a double patenting rejection under non-statutory grounds is proper.

It is respectfully noted that each and every claim of the present invention requires a "urine sample", while each and every claim of the co-pending application 11/770/372 requires a sample isolated from the blood, including a blood serum sample. Consequently, the claims of these applications cannot be the "same" invention, because there cannot be overlapping of the scope of the claims, and there cannot be any unjustly extending of the rights granted in a first patent issued to one of the applications, by the granting of the second application. The circumstances described in *In re Schneller*, 397 F.2d 350, 158 USPQ 210 (CCPA 1968), are not applicable in the present circumstances.

Applicants traverse, substantially on the same basis as argued above for co-pending Application No. 11/096,113; that is, that the secondary references utterly fail to teach, suggest or predict that the finding of NGAL in blood or serum following renal tubular cell injury would predictably include the use of urine as a "second window" of detection.

(iv) Co-pending Application No. 11/770,245 (Attorney Docket CHM-032BK)

Claims 2, 4-5, 9-11, 33, 35, 37, 55 and 66 are provisionally rejected on the ground of non-statutory obviousness-type double patenting over the claims 29, 31-43 and 45-49 of co-pending Application No. 11/770,245 (Applicants' commonly-owned, co-pending application, Attorney Docket CHM-032BK), in view of David et al., Ramsden et al., Blaser et al, Moses et al, either of Matthaeus 1 or Matthaeus 2 , and either one of Sanicola-Nadel or Muramatus et al.

Claim 60 is provisionally rejected on the ground of non-statutory obviousness-type double patenting over the claims 29, 31-43 and 45-49 of co-pending Application No. 11/770,245 (commonly-owned, co-pending application, Attorney Docket CHM-032BK), in view of David et al, Ramsden, Blaser, Moses, either of Matthaeus 1 or Matthaeus 2, either one of Sanicola-Nadel or Muramatus et al., and further in view of Zanardo et al.

Applicants request reconsideration and withdrawal of the provisional restriction requirement, on the basis that the claims of the present application are drawn to acute renal tubular cell injuries, whereas the claims of the reference application 11/770,245 are drawn to chronic renal tubular cell injuries. The acute and chronic injuries are generally caused by different types of events or diseases, and the Examiner has not identified any reference or line of reasoning which would demonstrate to a person of ordinary skill in the art that an effective biomarker for acute renal tubular cell injury would also be an effective biomarker for chronic renal tubular cell injury.

Applicants traverse the rejection on the basis that the co-pending claims specify a body fluid sample, while the instant claims utilize urine. The assertion of obviousness is based solely on Ramsden et al. (that urine is non-invasive), and that NGAL can be detected in urine (Blaser and Moses). Nonetheless, the factual findings pertaining to these references as a whole established earlier in this Response, makes it clear that the rejection fails to establish *prima facie* non-statutory obviousness-type double patenting, because the secondary references utterly fail to

teach, suggest or predict that the finding of NGAL in a body fluid generally following renal tubular cell injury would predictably include the use of urine as a "second window" of detection.

In any event, Applicants preserve the right to file a Terminal Disclaimer in the later of the present application and the reference Application No. 11/770,245 that has allowed claims.

CONCLUSION

Applicants believe a full and complete response to the Action has been made, and that the claims are patentable over the prior art of reference. Applicants request a prompt notice of allowance of the application.

Respectfully submitted,

For: Prasad DEVARAJAN et al.

By _____

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